INACTIVATION BY ULTRAVIOLET RADIATION

Bactericidal ultraviolet lamps radiate about 90 percent of their energy at 2537 A, a wavelength near 2650 A, the maximum for lethal activity. Ultraviolet penetrates only slightly below the surface of a liquid, and is primarily used to restrict microbial growth on surfaces and to sterilize air. Other than the indirect action of ozone produced in small quantities by these lamps, any shielding makes ultraviolet essentially ineffective unless a reflective surface is interposed.

Visible light of certain wavelengths applied after ultraviolet exposure can reverse the apparent ultraviolet inactivation. There is also evidence (157) that visible light can partially protect organisms subsequently exposed to ultraviolet. Ultraviolet can also increase sensitivity to heat, but this combination would be applicable only to surface sterilization under conditions of limited heat penetration within solids.

Perhaps the main use of ultraviolet would be in spacecraft assembly. Ultraviolet lamps in rooms or isolators used for spacecraft assembly will destroy airborne microorganisms provided they are not contained within dust particles or otherwise shielded. High intensity ultraviolet lamps in air conditioning ducts can destroy 90-99 percent of the organisms passing by the lamps.

Nagy (op. cit.) presented data necessary for calculating the lamp requirements for various size installations.

Federova (148) discussed the ultraviolet resistance of microorganisms in connection with protection which might be afforded by cosmic dust and the possibility of interplanetary transport of microorganisms. The ultraviolet resistance of microorganisms varies over a wide range of ultraviolet energy. Spores of bacteria and fungi are 10-40 times more resistant than vegetative cells. Organisms which produce dark spores are most resistant, but no organisms withstand about 2 X 10^3 erg/cm^2/sec longer than 3-4 min. Inactivation of 99 percent of the organisms in a closed room requires 1-1.5 X 10^6 erg/cm^2. Exposure to visible light prior to ultraviolet increases resistance to ultraviolet. A spore coated with dead cells or within a speck of dust will be somewhat protected because of the low penetration of ultraviolet.

In a recent publication, Nagy (American Industrial Hygiene Association Journal, v. 25, pp. 274-281, 1964) reviewed the properties and biological effects of ultraviolet generators. A useful table lists the energy required at 2537 A for 90 percent inhibition and for complete destruction of over 40 species of bacteria, yeasts, and molds. Aspergillus niger spores, the most resistant organism cited, required 132,000 microwatt-sec/sq cm for total destruction of colony forming capacity. Special lamps

emit a controlled amount of 1849 A radiation. This wavelength dissociates oxygen to produce ozone. Ozone is bactericidal at 0.04 ppm and 60-90 percent relative humidity.

Webb, Clark and Bailey (158) disclosed that large proportions of staphylococci will survive ultraviolet irradiation.

Cowling, Field and Alexander (Official Digest, v. 34, pp. 816-842, 1962) found that organic coatings considered for spacecraft temperature control are less rapidly degraded by ultraviolet radiation in a simulated space environment than in the normal terrestrial atmosphere.

Water Vapor.

Atmospheric water vapor absorbs much of the bactericidal ultraviolet. Most microorganisms are much more sensitive to ultraviolet when irradiated in a dry atmosphere or in vacuum.

Beebe (167) demonstrated that bacterial aerosols irradiated with bactericidal ultraviolet light are protected by water vapor in the atmosphere.

Luckiesh et al. (150) demonstrated that while certain organisms displayed increased resistance to ultraviolet with increasing humidity, others appeared less resistant at high humidities. Airborne mold spores were unaffected by wide variations in humidity. Aspergillus niger spores are notably resistant, requiring 1,800 and 9,000 microwatts/sq cm-min for 50 and 90 percent in-

activation in air, and 1,300 and 3,000 microwatts/sq cm-min for the same inactivation when irradiated on the surface of a culture medium. The high resistance of A. niger spores is attributed to the increase in free radicals in the spore melanin and darkening of melanin by ultraviolet.

Woodward et al. (152) studied the influence of water on ultraviolet sensitivity in several organisms. Neurospora conidia irradiated dry yielded about five times as many colonial mutants as conidia irradiated in water. Conidia were most sensitive to ultraviolet at or below 15 percent relative humidity. Bacteriophage XP4 was especially resistant to ultraviolet at 35 percent relative humidity, but in phage T_3, ultraviolet sensitivity was directly proportional to the degree of hydration.

Oxygen.

Zetterberg (159) could not detect any influence of oxygen on mutagenic or killing action of ultraviolet light exposure on Ophiostoma.

Oxygen protects T_1 bacteriophage against irradiation and hydrogen peroxide inactivation (91). The lethal effect of hydrogen peroxide and ultraviolet acting simultaneously was greater than the activity of each agent acting separately. Oxygen appears to be required for photoreactivation in Bacillus subtilis 168 (164).

Potentiation of Thermal Lethality.

Duggar and Anderson (124) reported that exposure of Saccharomyces cerevisiae to ultraviolet light followed by heat treatment at 50 C is 2-5 times more lethal than if the heat precedes ultraviolet irradiation.

Curran and Evans (76) showed that spores were more susceptible to heat after exposure to ultraviolet light. Bacillus cohaerens, the most heat resistant of three organisms studied, was most readily sensitized to heat by ultraviolet. Wavelengths in the 350-1,600 A range were more effective than 2537 A in inducing heat sensitization at 98 C.

Doudney (155) deduced that RNA and protein synthesis during the initial period of incubation following ultraviolet exposure is necessary for bacterial recovery but detrimental to subsequent recovery.

Photoreactivation.

Jaggar (157) observed that Escherichia coli B cells exposed to light of 3,100 to 6,000 A prior to exposure to low doses of 2537 A ultraviolet light were partially protected from ultraviolet.

Ultraviolet-inactivated Endamoeba histolytica achieved maximum photoreactivation with a 45 min treatment with visible light (156). Kelner (164) described experiments which indicated that Bacillus subtilis 168 requires oxygen for photoreactivation

after ultraviolet irradiation.

Modification of Ultraviolet Resistance.

According to Webb and Clark (151), successive ultraviolet irradiation of Nocardia corallina coccoids produced a significant increase in sensitivity.

Adler and Hardigree (165) found that mutation at the lon locus in Escherichia coli K-12 results in enhanced radiation sensitivity and several morphological changes. Another series of mutants (166) are more sensitive than the parent strain to ultraviolet, and differ from the lon mutant in several characters.

Nonlethal exposure to ultraviolet is known to produce significant numbers of mutants in some organisms. Mutational synergism has been shown by Shankel and Coupe (160) to greatly increase the number of mutants in Escherichia coli irradiated sublethally and then allowed to develop in the presence of caffeine. Alkaline conditions and incubation at 37 or 42 C favored the expression of mutants.

Ultraviolet resistance has been transferred in mating strains of Escherichia coli (161)

Payne and Campbell (162) determined that iron can protect Micrococcus violagabriellae from ultraviolet damage and possibly permit recovery from such damage. Subsequently (163), they obtained

evidence suggesting that a cytochrome-linked electron transfer system was ultraviolet sensitive.

A method for detecting ultraviolet damage producing respiratory deficiency was described by Pittman and Roshanmanesh (154).

Chemicals.

Bruni (149) noted that ultraviolet activated the oligodynamic action of silver.

Assessment of absolute lethality in any treatment involving ultraviolet should include a brief exposure of the material or organisms to visible light so that repairable cell damage can be detected.